

In re: Application of TERADA *et al.*
Serial No.: 10/045,721
Page 2 of 16

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. (Currently Amended) A method for identifying a drug candidate for promoting tissue-specific differentiation of a naïve-embryonic stem cell (ES), the method comprising the steps of:
 - (A) providing a library of test substances, the library comprising at least a first test substance and a second test substance, the first and second test substances having different molecular structures;
 - (B) providing an *in vitro* culture of undifferentiated embryonic stem cells (ES), the culture being divided into at least a first subculture and a second subculture and ~~the stem cell cultures~~;
 - (C) culturing the first and second subcultures stem cells for at least about 3 days in the absence of a test substance;
 - (D) contacting the first subculture with ~~the~~ a first test substance and the second subculture with the second test substance;
 - (E) culturing ~~for at least about 14 days~~, the first and second subcultures between 7 to 18 days wherein an early stage differentiation test substance is added on days 9 to 12, a mid stage differentiation test substance is added from day 12 to 18 respectively contacted with the first and second test substances under conditions that would promote tissue specific differentiation of the stem cells if an agent that promoted tissue specific differentiation was in contact with the subcultures stem cells; and
 - (F) analyzing the cells in the first and second subcultures for increased tissue-specific gene expression.
2. (Cancel).

{WP260227;1}

In re: Application of TERADA *et al.*
Serial No.: 10/045,721
Page 3 of 16

3. (Currently Amended) The method of claim [[2]] 1, wherein the embryonic stem cells are mammalian embryonic stems cells.

4. (Cancelled)

5. (Currently Amended) The method of claim [[4]] 1, wherein the murine embryonic stem cells are R1 embryonic stem cells.

6. (Currently Amended) The method of claim [[3]] 1, wherein the mammalian embryonic stem cells are human embryonic stem cells.

7. (Cancelled)

8. (Previously Presented) The method of claim 1, wherein the conditions that would promote tissue-specific differentiation of the stem cells comprises culturing the first and second subcultures at about 37°C in a humidified, carbon-dioxide containing incubator.

Claims 9-13. (Cancelled).

14. (Previously Presented) The method of claim 1, wherein the step (F) of analyzing the cells in the first and second subcultures for increased tissue-specific gene expression comprises isolating mRNA from the first and second subcultures.

15. (Original) The method of claim 14, wherein total cellular RNA is isolated from the first and second subcultures.

16. (Previously Presented) The method of claim 14, wherein the step (F) further comprises reverse-transcribing the mRNA to create cDNA.

{WP260227;1}

In re: Application of TERADA *et al.*
Serial No.: 10/045,721
Page 4 of 16

17. (Previously Presented) The method of claim 1, wherein the step (F) of analyzing the cells in the first and second subcultures for increased tissue-specific gene expression comprises performing a polymerase chain reaction (PCR).

18. (Original) The method of claim 14, wherein the isolated mRNA is immobilized on a substrate.

19. (Original) The method of claim 18, wherein the substrate is contacted with a probe that specifically hybridizes to the tissue-specific mRNA.

20. (Previously Presented) The method of claim 1, wherein the step (F) of analyzing the cells in the first and second subcultures for increased tissue-specific gene expression is performed using gene chip technology.

{WP260227:1}